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BIOSORPTION OF LONG-CHAIN FATTY ACIDS IN UASB TREATMENT PROCESS

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Abstract—Biosorption of long-chain fatty acids (LCFA) in the upflow anaerobic sludge blanket (UASB) treatment process was investigated using batch tests and continuous reactor runs. Batch experiments were conducted, for characterization of the biosorption, with two active and one inactivated (autoclaved) sludge granules as sorbents and with a single (oleic acid) or a mixture of LCFA ($LCFA_m$; 50% oleic, 35% palmitic and 15% stearic acid) as sorbates. The $LCFA_m$ showed a faster adsorption onto the granules than oleic acid. With the active sludge granules, adsorption was followed by desorption. Methane production increased significantly, either simultaneously (at lower LCFA concentrations) or succeedingly (at higher concentrations) with desorption. The desorption was mediated by biological activity, since it did not prevail with inactivated granules or with active granules inhibited at higher $LCFA_m$ concentrations. The inactivated granules had a slightly higher initial biosorption capacity. Increased LCFA concentrations resulted in more LCFA adsorbed and greater inhibition of their biodegradation. A hypothesis is proposed to explicate the relationship between biosorption, desorption and biodegradation of LCFA by sludge granules: adsorption is a prerequisite for biodegradation while desorption is a consequence of biodegradation. Isothermal studies with oleate showed that the apparent biosorption could be described by the physical multilayer adsorption theory and the sorption isotherm derived was consistent with the Freundlich model. The quantitative relation between LCFA biosorption and granular sludge flotation was investigated in a UASB reactor fed with $LCFA_m$. Sludge flotation depended on the $LCFA_m$ loading rate rather than on their concentration. The higher the loading implemented, the more flotation occurred and the shorter time required for complete flotation of the sludge bed. Flotation started when the $LCFA_m$ loading rate exceeded 0.09 g COD/g VSS·d, while complete flotation occurred at the loading rates exceeding 0.2 g COD/g VSS·d. These results suggest that sludge bed wash-out is likely to be encountered before inhibition of methanogenesis during the treatment of LCFA-containing wastewaters by the UASB process. © 1998 Elsevier Science Ltd. All rights reserved

Key words—biosorption, desorption, granular sludge, inhibition, isotherm, long-chain fatty acids, sludge flotation, UASB

INTRODUCTION

Wastewaters produced from edible oil refinery, slaughterhouse, wool scouring and dairy products industry contain a high (> 100 mg/l) concentration of lipids (characterized either as fats, oils or greases). In anaerobic wastewater treatment systems, lipids are readily hydrolyzed to long-chain fatty acids (LCFA) and glycerol, whilst the degradation (β-oxidation) of LCFA to acetate is regarded as the rate-limiting step (Novak and Carlson, 1970; Rinzema *et al.*, 1994). Moreover, LCFA are inhibitors of anaerobic microorganisms already at millimolar concentrations (Koster and Cramer, 1987; Hwu *et al.*, 1996). In addition, sludge flotation and/or wash-out in upflow anaerobic sludge blanket (UASB) reactors has been encountered following shock loads of milk fats (Samson *et al.*, 1985) and LCFA (Rinzema *et al.*, 1989). Adsorption of LCFA onto the surface of microbial cell was indicated as the mechanism of inhibition (Galbraith and Miller, 1973a,b), while that onto granular sludge was speculated as the reason for sludge flotation/wash-out (Lettinga and Hulshoff Pol, 1992).

Sorption of fatty matter or LCFA was reported as a relatively rapid process. Sorption equilibria of four LCFA sodium soaps are achieved after 30 min (Weatherburn *et al.*, 1950) or 16 h (Meader and Fries, 1952) of contact with textile fibers. Hrudey (1982) reported that 80% of the lipids are adsorbed to activated sludge within 20 min. For anaerobic sludge, Hanaki *et al.* (1981) found that LCFA disappear from the aqueous phase and accumulate in

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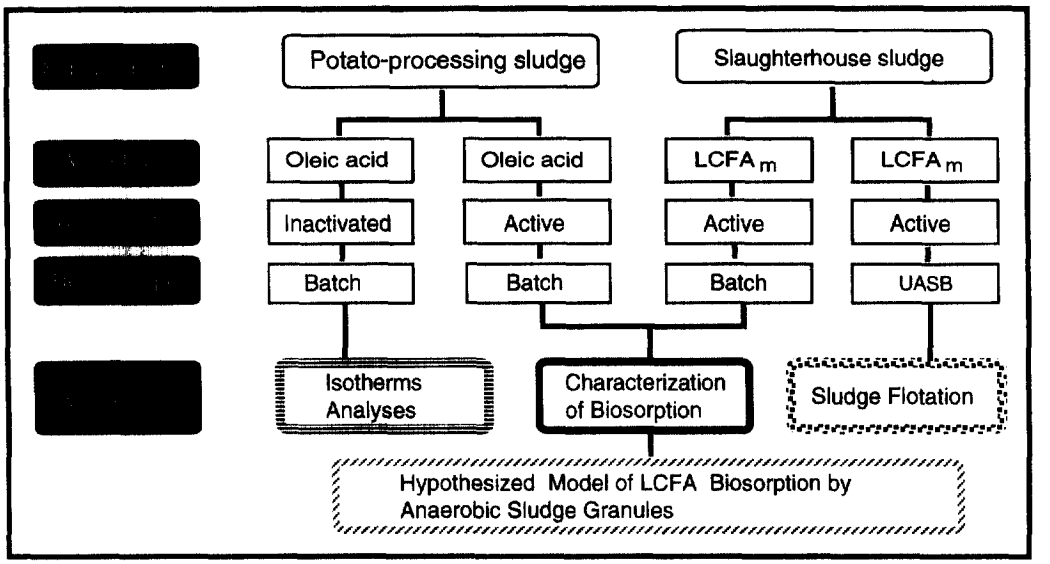

Fig. 1. Schematic diagram showing the experimental design protocol (detailed in text).

the solid phase within the first 24 h of incubation in a digester.

The present knowledge on adsorption and biodegradation of LCFA in UASB reactors is still limited. Adsorption of sodium salts of capric ($C_{10:0}$) and lauric acid ($C_{12:0}$) by methanogenic granular sludge was investigated, respectively, by Keurentjes and Rinzema (1986) and Koster (1987). However, these two LCFA are not present or, if any, in trace amount in raw materials (Taylor, 1965) and wastewaters (Viswanathan *et al.*, 1962). In fact, oleic acid ($C_{18:1}$) is the most abundant constituent in LCFA-containing wastewaters (Viswanathan *et al.*, 1962; Komatsu *et al.*, 1991; Quéméneur and Marty, 1994). Hence oleic acid was used as a model compound in the present study. In addition, a mixture of LCFA ($LCFA_m$), consisting of 35% palmitic ($C_{16:0}$), 15% stearic ($C_{18:0}$) and 50% oleic acid, was used to simulate the genuine composition of a local slaughterhouse wastewater (Yuan, 1995). This work was aimed to (i) perform isothermal studies using inactivated UASB granules, (ii) characterize the LCFA biosorption and (iii) determine the effect of biosorption on sludge flotation. Therefore mesophilic (40°C) batch incubations (apparent LCFA biosorption) and experiments with continuous flow UASB reactors (biosorption *vs.* flotation) were performed. The sorption theory was also approached by conducting isothermal sorption tests at 40°C with oleic acid by use of inactivated sludge granules.

MATERIALS AND METHODS

Assessing protocol

The experimental design used in this study is illustrated in Fig. 1. LCFA biosorption by anaerobic sludge granules was assessed in batch and continuous reactors using two origins of anaerobic granular sludge with either oleic acid or the $LCFA_m$ as sorbate.

Anaerobic granular sludges

Granular sludge taken from a 110 m^3 internal circulation reactor treating potato processing wastewater (Agrico, Wezep) was elutriated and well settled. Size distribution analyses, performed as described previously (Hwu *et al.*, 1996), showed that 87% of the granules had a size between 0.5–2.0 mm in diameter. Active sludge granules were used in the experiments with oleic acid as the sole sorbate. For isothermal sorption tests, these granules were inactivated by autoclaving (121°C, 1.5 bar) for 1 h, followed by a second autoclaving (15 min) on the next day. This treatment was the only one with a nearly 100% inhibitory effect. Other chemical (*e.g.*, $HgCl_2$, NaN_3 and H_2O_2) or physical (gamma-ray irradiation) inactivation procedures did not completely inactivate the granules (unpublished data). For the experiments with $LCFA_m$ as sorbate, sludge granules from a slaughterhouse wastewater treatment plant (Da-Ann, Taipei) were used after elutriation and settling. These granules, examined by microscopy (Nikon Optiphot, Japan), had a size typically between 0.5–1.0 mm in diameter.

Media

Sodium oleate and a mixture ($LCFA_m$) of sodium salts of 35% palmitic, 15% stearic and 50% oleic acid (on the basis of chemical oxygen demand, COD) were used as sorbates. The $LCFA_m$ simulated the 3 major constituents in a local slaughterhouse wastewater, which represents at least 80% of the total lipid COD. To obtain a compatible ionic strength, solutions of mineral nutrients and trace elements (Hwu *et al.*, 1996) were also provided in experiments with inactivated sludge (isotherm analyses). Media were buffered by $NaHCO_3$ solution (5 g/l). After the addition of sorbates, the pH was immediately adjusted to 7.2 ($\pm$0.1) by adding drops of HCl.

Batch sorption tests

Biosorption of oleic acid and $LCFA_m$ was characterized and isotherm analyses were performed with batch-type reactors under anaerobic conditions (Fig. 1). Experiments

with single LCFA were done in glass serum bottles (136 ± 1 ml) with inactivated or active granular sludge (final total solids (TS) concentration of 2.5 g/l), supplemented with oleate of various concentrations (300, 600, 1000, 1400 and 2000 mg/l). The liquid volume (incl. sludge) in the bottle was about 70 ml. Subsequently, the headspace of the bottles was flushed with N_2/CO_2 gas (70/30, v/v) for 5 min. Bottles were then placed in a temperature controlled (40°C) reciprocating water-bath shaker (approximately 50 strokes/min). For the isotherms analyses, the Freundlich and the Langmuir models were considered. Upon finishing experiments, the appearance of the granules was studied by an Olympus ZS40 zoom microscope. Experiments with $LCFA_m$ were done with approximately 50 ml liquid (incl. 2.4 g TS/l) supplemented with $LCFA_m$ (150, 300, 600, 1000 or 1500 mg/l) in a 125(±1) ml glass serum bottle, incubated in a temperature controlled (35°C) rotary water-bath shaker (approximately 120 rpm).

UASB continuous reactor runs

The Plexiglas cylindrical UASB (height 850 mm, width 100 mm) was used to investigate the quantitative relation between LCFA biosorption and sludge flotation. The reactor had a working volume of 6.67 l and was temperature controlled at 35°C by a water jacket. The reactor was seeded with the same sludge used in $LCFA_m$ batch tests and had an initial biomass concentration of 37.5 g volatile suspended solids (VSS) per liter. During reactor start-up, glucose (2 g COD/l) was used as the carbon and energy source. When 80% COD removal efficiency was stably reached over 5 days, the reactor was supplied the same medium used in the $LCFA_m$ batch tests. Adjustment to various loading rates (from 0.086 to 0.250 g $LCFA_m$-COD/g VSS·d, in arbitrary order) was obtained by altering $LCFA_m$ concentrations and/or hydraulic retention times (HRT). The liquid superficial upflow velocity (V_{up}) was 2 m/h throughout the study. The initial volume of the sludge bed in each test run was recorded. Sludge flotation was monitored by visual observation and the time was noted when sludge started to float from the sludge bed. A test run was assumably finished either when flotation became trivial or when the entire sludge bed floated. In the latter case, the time period from initial to entire floating was recorded. Between two test loadings, floated matter, if any, was collected and gently stirred to remove glutinous materials and adhered biogas bubbles. This procedure allowed sludge granules to re-settle down. After reintroducing the sludge into the UASB, anoxic tap water was pumped through the reactor until no biogas production was detectable.

Analyses

In batch tests using oleate, samples from the supernatant were taken at certain time intervals and analyzed for their oleic acid content. If not analyzed immediately, samples were acidified below pH 2 and stored at −18°C. Samples (0.5 ml) were acidified with 2 drops of 6 N HCl and subsequently extracted with 5 ml petroleum ether (boiling point 40–60°C). A known concentration of sodium oleate was also analyzed in parallel as a positive control. After 1 h mixing, the ether phase was transferred to another tube using a Pasteur pipette. Ether was entirely evaporated by heating the tube in an 80°C water bath. Methylation reagent (1 ml) composed of fuming HCl and dry methanol (1:19, v/v) was added, well-mixed and then placed back in the 80°C water bath. After 30 min esterification, the tube containing methyl esters was cooled down to room temperature. Subsequently, 2 ml petroleum ether was added and vortexed for 3 min. One ml of this ether phase was Pasteur pipetted into a glass vial and immediately capped. The oleic (or other LCFA, from capric to arachidic, $C_{20:0}$) concentration in the vial was determined by a gas chromatograph (HP 5890 II) equipped with an auto sampler and a flame ionization detector. The column (25 m × 0.25 mm) used was coated with CP-WAX-58 (film thickness: 0.2 mm). Temperature conditions were: injector, 275°C; detector, 250°C; oven, programmed 140–240°C at 5°C/min. Helium was used as carrier gas with a flow rate of 1.1 ml/min. In each analysis, a fatty acids methyl esters (FAME) standard was also analyzed, which was used to identify and quantify the LCFA in the samples. Over 90% of the recovery for the positive control was usually found. The errors of the FAME standard between each analysis were less than 3%. COD was determined for samples from the experiments using $LCFA_m$. Otherwise stated, COD values were converted and expressed as mg $LCFA_m$/l based on the calculation that each gram of $LCFA_m$ theoretically equaled 2.893 g COD. The methane content was measured by gas chromatography (Hwu *et al.*, 1996). TS, VSS and COD determinations followed the procedures described in the standard methods (APHA, 1992).

Chemicals

Sodium salts of LCFA and FAME standard (AOCS No. 5) were purchased from Sigma, U.S.A. All chemicals were of analytical grade.

RESULTS AND DISCUSSION

Characterization and isotherm of oleate biosorption

In the oleate biosorption tests, oleic acid methyl ester was the main constituent among all LCFA peaks shown in the GC-chromatograms. No other intermediates higher than capric could be detected or, if any, in trifling concentrations. The profiles of residual oleate concentrations (ROC) in the aqueous phase and methane production from oleate conversion are presented in Fig. 2. Active sludge granules had a different sorption behavior from inactivated sludge granules. In general, the inactivated granules showed a slightly higher initial biosorption capacity for oleate as indicated by the lower ROC after 1 d of incubation. This might be due to the alteration of the surface properties of sludge granules by autoclaving, as was pointed out by Tsezos and Bell (1989) that surface properties of microbial cells will change after their death. With respect to the overall biosorption, however, the variation may not be large enough to have a significant difference between inactivated and active granules. This agrees with Ning *et al.* (1996), who postulated that anaerobic biosorption is mainly a physical-chemical process. The present work further confine biosorption of LCFA merely to a physical process (see below).

Oleate was adsorbed prior to being biodegraded since active granules removed 40–70% of oleate from the aqueous phase in the first day; meanwhile, less than 1% of methane (on COD basis) was produced. This indicates that the primary mechanism for COD removal of LCFA is biosorption rather than biodegradation, as was also found for synthetic milk substrate with anaerobic granules (Riffat and Dague, 1995). Figure 2 further shows that a desorption phenomenon (increasing ROC) was

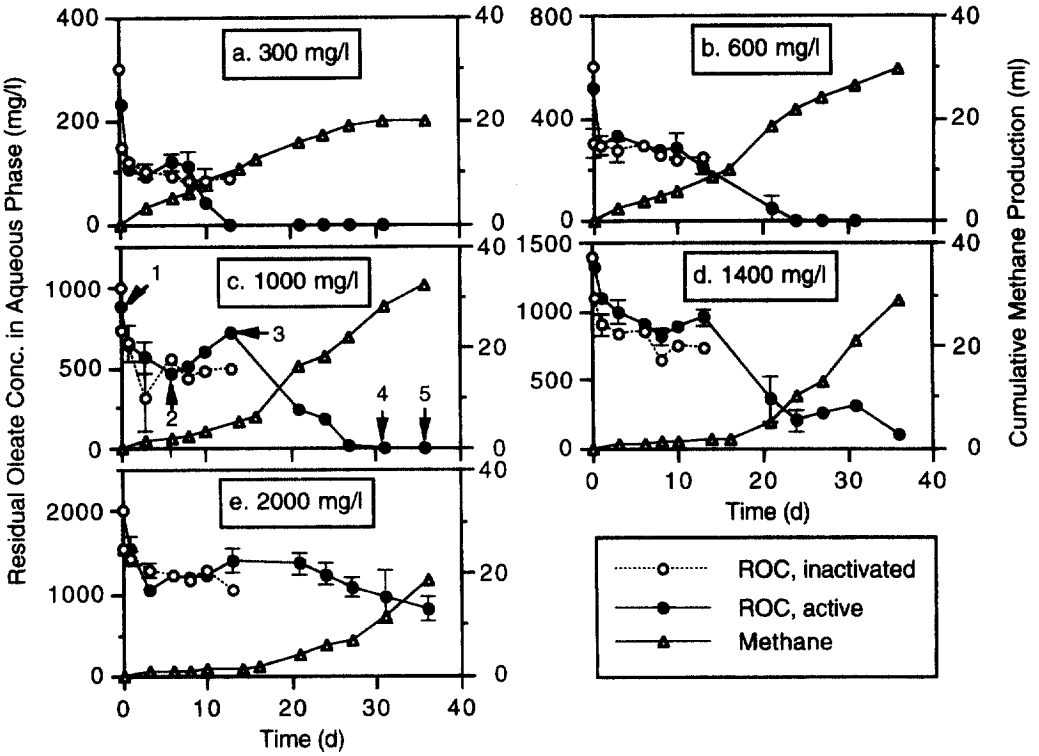

Fig. 2. Profiles of cumulative methane production by active sludge granules and residual oleate concentrations (ROC) in aqueous phase with active and inactivated sludge granules. Numbered arrows in graph (c) indicate data taken for mass balance calculation at their corresponding time points [Fig. 4(a)]. Bars indicate standard deviations (n = 3).

commonly found in each concentration after the first adsorption (decreasing ROC). Unlike the active granular sludge, no clear desorption was observed with the inactivated granular sludge, suggesting that the desorption was biologically mediated. This finding is in agreement with Tsezos and Bell (1989), who attributed the desorption from live microbial biomass to the biodegradation of organic molecules. Indeed, the desorption was accompanied by a significant increase of methane production, either simultaneously (at oleate concentrations of 300 and 600 mg/l) or succeedingly (1000 mg/l and higher). Methane production started after a lag period at oleate concentrations higher than 600 mg/l, which agreed well with the oleate toxicity test (IC_{50} = 612 mg/l) performed with the same granules at the same temperature (Hwu *et al.*, 1996). Moreover, oleate adsorption was significantly concentration dependent, viz., the higher the initial oleate concentration added, the more oleate adsorbed, corresponding with a higher methanogenic inhibition (*i.e.*, a longer lag period of methane production). This agrees with Galbraith and Miller (1973a,b), who indicated that LCFA adsorption onto the surface of microbial cell is the mechanism of inhibition.

Following the desorption phenomenon, ROC decreased again to concentrations lower than during the first adsorption (Fig. 2). This decrease was no longer completely due to oleate adsorption, but high-rate methane production partly contributed to the decrease of ROC as well. Methane production did not stop, but further increased when the ROC had decreased below the detection limit (20 mg/l) for 7 days [Fig. 2(a–c)]. This indicated that the adsorbed oleate on the surface of sludge granules was gradually biodegraded without detectable desorption. Upon termination of the experiment, a complete biodegradation was observed merely at an oleate concentration of 300 mg/l [Fig. 2(a)]. However, it can be expected that oleate with all concentrations tested would eventually convert to methane.

The adsorption rates of oleate by anaerobic sludge granules were rather slow (in h^{-1} or d^{-1} scale) compared with those of oleate/LCFA soaps by textile fibers, where adsorption occurred at a min^{-1} scale (Weatherburn *et al.*, 1950; Meader and Fries, 1952). This difference can be attributed to the larger specific surface area of textile fibers compared with sludge granules. While conducting sorption tests with soaps, critical micelle concentrations (CMC) of soaps play a determinative role in the state of reaction. Adsorption phenomena become dominant when soap concentrations are increasingly approaching their CMC levels. The oleate concen-

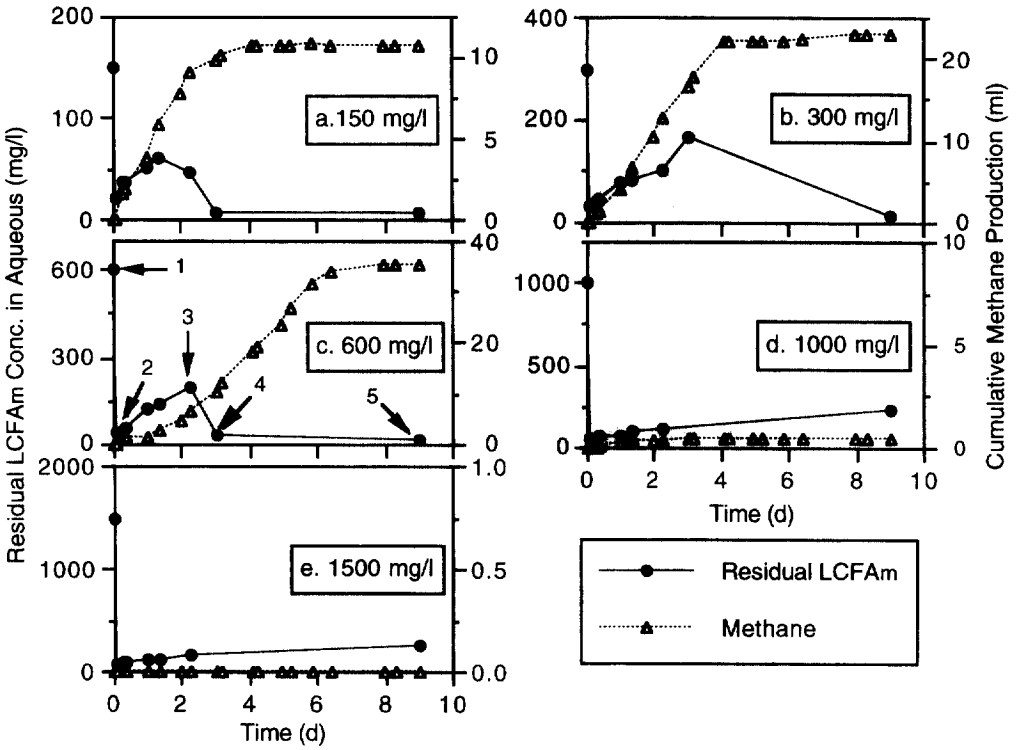

Fig. 3. Changes of methane production and residual $LCFA_m$ concentrations. Numbered arrows in graph (c) indicate data taken for mass balance calculation at their corresponding time points [Fig. 4(b)].

trations (0.99–6.57 mM) used in this work were much higher than its CMC (0.56 mM; calculated as described by Gerrens and Hirsch, 1974). Moreover, the oleate concentrations were also much higher than those of bivalent cations in solution, so that oleate precipitation was negligible in our experiments. Thus oleate sorption by anaerobic sludge granules is a relatively long-term process. This is in agreement with adsorption of capric acid by inactivated anaerobic sludge granules, where no equilibrium could be reached after 8 days (Keurentjes and Rinzema, 1986). In contrast, lauric acid rapidly disappears from the aqueous phase when incubated with these granules, which is attributed to precipitation (Koster, 1987).

Pseudo-equilibria of oleate adsorption by the inactivated granules were presumed on day 13 (Fig. 2). The data were analyzed for isotherms according to the Freundlich and the Langmuir models. Of the two isotherm theories considered, the Freundlich model gave the highest correlation ($r^2 = 0.984$), with a K = 12 mg/g and $1/n$ = 0.521:

$$q = KC^{1/n}$$

where q = equilibrium amount of adsorbate on adsorbent (mg oleate/g TS); C = equilibrium concentration in aqueous phase (mg oleate/l); and K, $1/n$ = Freundlich parameters.

Hence, biosorption of oleate by anaerobic granules can be regarded as a physical multilayers adsorption, in contrast to that of soaps by textile fibers which was described as a chemical adsorption (Weatherburn *et al.*, 1950).

The appearance of granules was examined upon termination of this experiment (day 36). The oleate adsorption was visually evidenced. More "white granules" were observed at higher oleate concentrations, probably due to a coating of adsorbed oleate. Oleate was not equally distributed (adsorbed) to each granule, although less distinction was found with inactivated granules. The variation between individual granules can be attributed to different sizes, surface properties and metabolic activities (biodegradation of oleate by active granules).

Characterization of $LCFA_m$ biosorption

Figure 3 shows that $LCFA_m$ biosorption had a trend similar to oleate biosorption (Fig. 2). Methane production with longer lag periods prevailed at increasing $LCFA_m$ concentrations and became almost negligible at the two highest concentrations, suggesting inhibition of methanogenesis. Adsorption of $LCFA_m$ proceeded much faster than that of oleate by anaerobic sludge granules. More than 90% of the $LCFA_m$ was removed from the aqueous phase within the first 3 h, while it took at least 24 h before 40–60% of the oleate was removed (Fig. 2). Sorption of LCFA soaps by textile fibers at 30 and 50°C has no significant differences (Weatherburn *et al.*, 1950). Therefore the influence

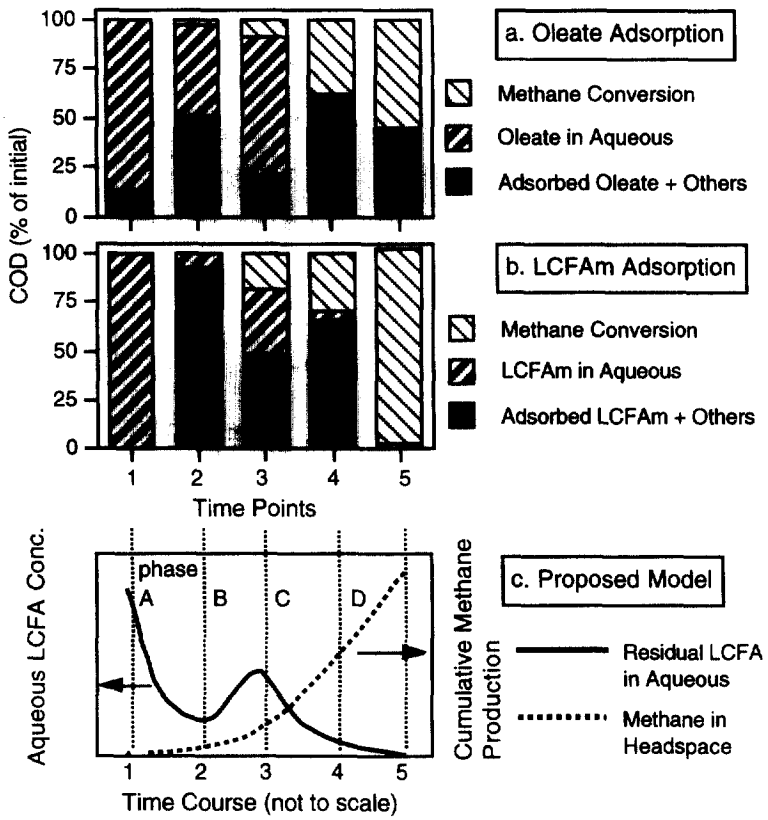

Fig. 4. Mass balance diagrams of (a) oleate and (b) $LCFA_m$ biosorption tests at 1000 and 600 mg/l initial concentrations, respectively. Time points refer to Fig. 2(c) and Fig. 3(c), respectively. The four-phase model (c) describing LCFA biosorption by anaerobic sludge granules.

of temperature difference (merely 5°) in the present work can be expected to be of minor importance. Apart from the complex nature of LCFA, the different behavior between oleate and $LCFA_m$ biosorption tests can be attributed to differences in sludge origin (Ning *et al.*, 1996) or granular size (Riffat and Dague, 1995).

Assuming a linear relationship between time 0 and 2.28 h (the first two data points of the $LCFA_m$ curves, Fig. 3), the initial adsorption rates of $LCFA_m$ amount to 0.023, 0.027, 0.032, 0.045 and 0.065 h^{-1} for concentrations of 150, 300, 600, 1000 and 1500 mg/l, respectively. These results clearly show that increasing LCFA concentrations resulted in increasing initial adsorption rates. Like oleate biosorption, $LCFA_m$ biosorption was also a concentration dependent reaction. Within nearly 10 days neither desorption nor significant methane production was observed when $LCFA_m$ concentrations exceeded 1000 mg/l. These results are in agreement with those of oleate biosorption by inactivated sludge and further indicate that the desorption of LCFA was mediated by biological activity.

Similar to the result of oleate biosorption test at 300 mg/l concentration [Fig. 2(a)], complete biodegradation was attained at the two lowest imposed concentrations of 150 and 300 mg $LCFA_m$/l [Fig. 3(a, b)]. In contrast, mass balance calculations (data not shown) indicate that the amount of adsorbed $LCFA_m$ was already zero before the start of the two second declines of residual $LCFA_m$ concentrations in the aqueous phase. Thus the second decrease can not be attributed to adsorption but to a prevailing methane conversion of $LCFA_m$. However, the second decrease at 600 mg $LCFA_m$/l [Fig. 3(c)] was indeed due to the occurrence of both adsorption and methane conversion, as was also found for all oleate biosorption tests. Compared to Fig. 2(a and b), the shorter time period required for complete degradation at $LCFA_m$ concentrations below 600 mg/l might be due to the fact that the sludge used was acclimated to slaughterhouse wastewater.

Model of LCFA biosorption by anaerobic sludge granules

Both big similarities and distinct differences were found between oleate and the $LCFA_m$ adsorption. The LCFA biosorption therefore clearly can be regarded as a very complex behavior. To generalize the biosorption behavior, data at some important time points in Fig. 2(c) and Fig. 3(c) (indicated by arrows with Arabic numbers) were converted to their COD balance diagrams, respectively, shown in

Table 1. Sludge flotation in UASB reactors exposed to LCFA

Compound investigated	Reactor Vol. (l)	Temp. (°C)	Dimensions (mm) height	width	HRT (h)	V_{up} (m/h)	LCFA loading rate (g COD/g VSS·d)	Concentration (mg LCFA/l)	Degree of sludge flotation (%)[a] (time[b])	Data source
$LCFA_m$	6.67	35	850	100	27.6	2.0	0.086	192	0	This study
					27.6		0.106	236	19	
					27.7		0.173	387	74	
					27.6		0.222	495	100 (97)	
					27.9		0.250	565	100 (93)	
					16.0		0.120	155	43	
					17.5		0.144	203	68	
					16.0		0.203	263	100 (107)	
Lauric acid	0.20	30	170	39	1.6	0.1	0.059	102	0	adapted
							0.118	205	100 (7)	from
							0.177	307	100 (4)	Rinzema
							0.235	409	100 (3)	*et al.*
							0.294	511	100 (3)	(1989)

[a]Degree of sludge flotation was defined as the ratio of total amount of floated sludge (ml) to the total amount of inoculated sludge (ml).
[b]In parentheses, time (h) required for complete sludge flotation.

Fig. 4(a and b). Based on the trends shown in Fig. 4(a and b), a model is proposed to idealize profiles of cumulative methane production and residual LCFA concentration [Fig. 4(c)]. Being introduced to a reactor inoculated with granular sludge, LCFA dramatically disappear from aqueous phase and adsorb onto solid phase [phase A, Fig. 4(c)], presumably increasingly in a multilayer at higher LCFA concentrations. At this phase no significant methane production occurs, probably due to a response to LCFA toxicity. Since LCFA adsorption is a concentration dependent reaction, the higher concentration will lead to the larger amount adsorbed and, consequently, the longer lag period of methanation. Subsequently, LCFA concentration in aqueous phase increases, indicating the occurrence of desorption (phase B). However, it will not be the case if methanogenesis is seriously inhibited. After the desorption, a second dramatic disappearance of LCFA from aqueous phase occurs (phase C). In general, a significant methane production can be observed during this phase. Depending on the LCFA concentration imposed, this phenomenon can be attributed to a second adsorption (at high concentrations) or a good biodegradation (at low concentrations). Finally, biodegradation of LCFA continues and all the adsorbed LCFA is eventually converted to methane (phase D). It has to be noted that this model was derived from biosorption by active granules only, as such a model can be applied to simulate anaerobic treatment of LCFA in bioreactors.

In Fig. 4(a and b), COD recoveries from the biomass yield are not shown because kinetic parameters available in the literature (Novak and Carlson, 1970) are, in the present point of view, not applicable since they were derived from the evolution of the COD removal in kinetic calculations. The proposed model agrees with Sayed *et al.* (1988), who reported that adsorption is the mechanism involved in the COD removal of slaughterhouse wastewater. The above-mentioned model [Fig. 4(c)] further indicates that the quickly and largely initial COD removal does not necessarily imply biodegradation. Hence, when LCFA removal is monitored by COD, TOC (total organic carbon) or DOC (dissolved organic carbon), LCFA bioconversion might be overestimated. Accurate description of the anaerobic conversion of LCFA requires also information on their metabolites. The final metabolite, *i.e.*, methane, is an elegant and easily detectable indicator for biological activity on LCFA and therefore differentiates the substrate removal by adsorption only.

Effect of $LCFA_m$ biosorption on UASB treatment process

To simulate the LCFA biosorption in a practical treatment process, 8 test runs were performed with a UASB reactor fed with $LCFA_m$. Table 1 and Fig. 5 compare the experimental conditions and results from the present study with a previous study (Rinzema *et al.*, 1989). Based on (1) the different biosorption characteristics between oleate and $LCFA_m$ described earlier in this article and (2) the higher V_{up} may lead to greater sludge flotation/wash-out; one might expect a more serious sludge flotation occurring in the present study. On the contrary, however, Rinzema *et al.* (1989) observed more severe flotation. Apart from the often encountered "piston effect" (experimentally easily caused by laboratory scale reactors with small diameters), the very short HRT applied in their study (1.6 h) might be another possible explanation. In our case, the $LCFA_m$ clearly led to serious sludge flotation, although good COD removal efficiencies (82–93%) were achieved under all loadings tested. Despite the adsorptive removal, such high efficiencies can not be guaranteed in practice, as devices to retain sludge in laboratory scale reactors are usually not applicable for industrial scale treatment plants.

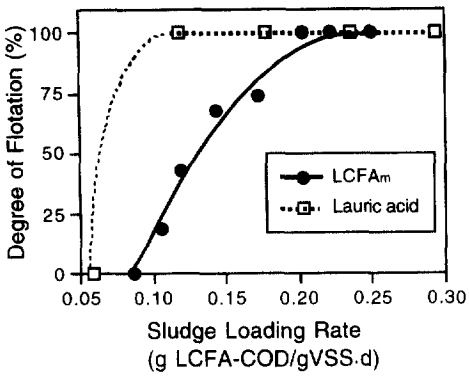

Fig. 5. Relation between sludge loading rate and sludge flotation in UASB reactors treating $LCFA_m$ (this study) and lauric acid (adapted from Rinzema *et al.*, 1989).

The continuous experiments showed that the higher the applied loading rates of $LCFA_m$, the more flotation occurred and the shorter time required for a complete flotation (Fig. 5 and Table 1). In contrast to batch-type biosorption experiments, sludge flotation depended upon LCFA loading rates rather than on LCFA concentrations. Obviously reactor hydrodynamics resulted in this difference. Flotation started when the $LCFA_m$ loading rate exceeded 0.086 g COD/g VSS·d while complete flotation occurred at loading rates higher than 0.203 g COD/g VSS·d. Samson *et al.* (1985) reported that sludge flotation results in the treatment failure of an industrial scale UASB reactor treating milk fat (whose major hydrolysis product is oleic acid). Lettinga and Hulshoff Pol (1992) suggested that adsorption of fatty matter on sludge particles leads to sludge flotation, which agrees with this work. In this study, a quantitative relation between $LCFA_m$ biosorption and sludge flotation was derived (Fig. 5). This relation is, to the best of our knowledge, the first one reported for the anaerobic treatment of wastewaters containing fatty matter or their main ingredients.

It should be noted that even at 0.203 g COD/g VSS·d condition, the corresponding $LCFA_m$ concentration (263 mg/l) was far below the minimum inhibition concentration (401 mg $LCFA_m$/l; Yuan, 1995) of methanogenesis. This suggests that, under practical conditions, complete sludge bed wash-out is likely to be encountered prior to inhibition of methanogens. Hence, biosorption of LCFA by anaerobic sludge granules significantly affects more sludge flotation than inhibition of the methanogenic consortia. Thus more research towards bioreactor systems that minimize adsorption, *e.g.*, by maximizing biodegradation (Hwu *et al.*, 1997a), or that are not affected by sludge flotation, *e.g.*, recirculation of wash-out biomass (Hwu *et al.*, 1997b), is required.

CONCLUSIONS

(1) Adsorption of LCFA by anaerobic sludge granules can be described by the Freundlich model. The initial adsorption rate is somewhat fast (1 day), but adsorption equilibrium is a relatively slow process (weeks). Desorption was only observed with active methanogenic granules, suggesting that the phenomenon is due to a biologically mediated reaction.

(2) A hypothesized four-phase model describing the behavior of LCFA biosorption by anaerobic sludge granules is proposed to simulate the practical treatment process.

(3) Both LCFA adsorption and toxicity to anaerobic granular sludge are concentration dependent.

(4) Sludge floatation is caused by adsorption and depends more on LCFA loading rates rather than on LCFA concentrations. LCFA biosorption can impede the success of a high-rate anaerobic wastewater treatment system such as a UASB reactor. Deterioration of the UASB treatment process by biosorption of LCFA is mainly due to sludge flotation rather than to the intoxication of the methanogenic consortia.

(5) Adsorption of LCFA is prerequisite for their biodegradation. However, an excessive adsorption can result in inhibition of methanogenesis and, more importantly, flotation of biomass.

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